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Synthesis of a cellulose thiosulfate and its immobilization on gold surfaces

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Abstract

A new water soluble cellulose derivative 6-*O*-(2,3-bis(thiosulfato)propyl-oxy-2-hydroxy-propyl)-cellulose (TSHP) was synthesized by addition of tetrathionate to 6-*O*-(allyl-oxy-2-hydroxy-propyl)-cellulose (AHP). On gold surfaces, TSHP forms dense monolayers by chemisorption with a mean thickness of 4 ± 1 nm. These monolayers were investigated by means of null-ellipsometry, AFM, FTIR-ERS, X-ray photoelectron spectroscopy and contact angle measurements. Based on the XPS results, it was proposed that the thiosulfato groups attached to the cellulose chains are homolytically cleaved and the resulting thio radicals bind covalently to the gold substrate. The formation of gold thiolates leads to a stable TSHP monolayer on the gold surface. The interaction between the TSHP films and fibrinogen and bovine serum albumin was also studied. The experimental results showed that the non-specific adsorption of these proteins was significantly reduced by the coverage of gold surfaces with TSHP.

Keywords: Gold; Cellulose; Monolayers

1. Introduction

The modification of metallic surfaces is of interest both academically and for its technological applications. Polymer coatings on metals can be very useful in the protection of microelectronics, in the stabilization of colloidal dispersions [1] and in the development of biocompatible materials, for example [2].

Ultrathin coatings of metals (e.g. gold, silver) can be obtained by the chemisorption of alkane thiols [2–5] and disulfides [6–8]. The sulfur atoms are covalently bonded to the metal surfaces, resulting in quasi-crystalline hydrophobic monolayers. Monolayers of hydrophilic compounds (e.g. hydroxy-alkyl thiols [9,10], cyclodextrin thiols [11–13]) have also been assembled on gold surfaces. Although monomeric monolayers are important to the understanding of microstructural properties, they are not sufficiently stable or durable for some applications [14]. A great deal of work [15–21] has therefore been devoted to the formation of ultrathin self-assembled polymeric films on solid surfaces. Polymers carrying chemically reactive groups have an advantage over monomers in that they can attach to the substrate at numerous points, yielding a more robust monolayer. For example, polystyrene bearing thiol end groups [20], styrene-propylene sulfide block copolymers [20], copolymers consisting of styrene and (mercaptomethyl) styrene units [21], sulfur-derivatized polyacrylates [18] and polymethacrylates [16,17] and grafted polysiloxane copolymers with alkyl disulfide and perfluoroalkyl side chains [15] all adsorb irreversibly onto gold and give highly stable monolayers.

In this paper we report the synthesis and chemisorption onto gold of a thiofunctionalized cellulose. 6-*O*-(2,3-bis(thiosulfato)propyl-oxy-2-hydroxy-propyl)-cellulose (TSHP) has the advantage of being water-soluble and readily available. The structure of the resulting layers was investigated by ellipsometry, contact angle, FTIR-ERS, XPS and AFM. A model to explain the adsorption mechanism of THSP is proposed. The influence of the cellulosic layers on the adsorption of fibrinogen and bovine serum albumin was also studied, using AFM and FTIR-ERS.

2. Experimental section

2.1. Synthesis

The synthesis was performed in two-steps: first cellulose was derivatized to 6-*O*-(allyl-oxy-2-hydroxy-propyl)-cellulose

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(AHP), then AHP was converted to 6-*O*-(2,3-bis(thiosulfato)-propyl-oxy-2-hydroxy-propyl)-cellulose (TSHP), as described below. NMR spectra were recorded on a Varian VXR-300 spectrometer (^{1}H:299.969 MHz, ^{13}C: 100.62 MHz) and FTIR spectra on a Bruker IFS 28 (Karlsruhe, Germany) using the DRIFT technique.

The molecular weight was determined by gel permeation chromatography (columns Suprema 1000, Suprema 30 and Suprema 3000; eluent 0.1 M NaCl) using a VISCOTEK (Houston, TX) differential viscometer at 35°C and a universal calibration with pullulane standards.

2.2. 6-O-(Allyl-oxy-2-hydroxy-propyl)-cellulose (AHP)

Five grams (30.9 mmol) cellulose (Avicel, PH101, Fluka) was stirred in 90 ml 25% NaOH solution at r.t. for 2 days. After dilution with 90 ml water, 36.8 ml (310 mmol) allyl glycidyl ether (Aldrich) was added dropwise at r.t. under N_2. The temperature was then raised to 60°C. After 6 days the mixture was cooled to r.t. and neutralized with conc. HCl. The aqueous solution (100 ml) of the resulting crude product was ultrafiltered against a Nadir membrane (Hoechst, type UF-PA-5, molecular weight cut-off 5000 g/mol) by 700 ml water. The retentate was freeze dried to yield 3.15 g (80%) AHP as a white solid. ^{1}H-NMR (D_2O): δ 5.78 (m, 0.7 H, vinyl), 5.08–5.20 (m, 1.4 H, vinyl), 4.34 (s, 1 H, H-1), 3.90 (s, 1.2 H, CH_2 allyl), 3.82 (m, 0.6 H, glycidyl), 3.64 (m, 2 H, H-6), 3.40–3.39 (m, 3 H, H-3, H-4, H-5), 3.37 (d, 2.4 H, glycidyl), 3.05 (m, 1 H, H-2); IR: 3399s, 2871s, 1644m, 1419w, 1369w, 1316w, 1250w, 1066s, 932w cm^{-1}; Anal. ($[C_6H_{10}O_5]_{0.3}[C_{12}H_{20}O_7]_{0.7}$) C, H: calc. C: 43.48, H: 7.66; found, C: 43.39, H: 6.46.

2.3. 6-O-(2,3-bis(thiosulfato)propyl-oxy-2-hydroxy-propyl)-cellulose (TSHP)

Potassium tetrathionate (3.18 g; Aldrich) was added to a solution of 1 g AHP in 100 ml water, which was then refluxed for 2 days. After cooling to r.t., the solution was filtered through a paper filter. The clear filtrate (100 ml) was then ultrafiltered against a Nadir membrane (see above) using 500 ml water. The retentate was freeze dried to give 0.44 g (34%) TSHP as a white solid. ^{1}H-NMR (D_2O): δ 5.76 (m, 0.6 H, vinyl), 5.08–5.20 (m, 1.2 H, vinyl), 4.34 (s, 1 H, H-1), 3.89 (s, 1 H, CH_2 allyl), 3.82 (m, 2 H, H-5, H-4), 3.45–3.17 (m, 9 H, H-2, H-3, H-6, glycidyl, thiosulfatopropyl); IR: 3399s, 2871s, 1644m, 1419w, 1369w, 1316w, 1250w, 1198m, 1017m, 932w, 601s, 408m cm^{-1}; weight average molecular weight M_w = 51 200 g/mol (by GPC); Anal. ($[C_6H_{10}O_5]_{0.3}[C_{12}H_{20}S_4O_{13}]_{0.1}[C_{12}H_{20}O_7]_{0.6}$) C, H, S: calc. C: 41.65, H: 6.72, S: 4.35; found, C: 41.1, H: 6.35, S: 4.57.

2.4. Gold substrates

A 40–50 nm layer of gold was thermally evaporated onto silicon wafers (2 × 2 cm^2, CrysTec Berlin, covered with 2 nm SiO_2 cleaned by a standard method [22] and wiped with isopropanol) at a pressure of 10^{-5} mbar. The coated wafers were then kept in a desiccator until use. The gold films were 50 ± 5 nm thick, the exact values in each case determined by null-ellipsometry (for details, see Section 2.7).

2.5. Immobilization of TSHP on gold

The gold covered silicon wafers were immersed in the aqueous TSHP (concentration 0.2–3 mg/ml) at ambient temperature (20–25°C) for 24 h. They were then washed 10 times with water and dried under a stream of N_2.

2.6. Adsorption of blood proteins

After characterization the TSHP coated gold was immersed for 24 h at r.t. in 1.0 mg/ml aqueous solutions of bovine serum albumin (BSA) factor V (Merck, Germany) or bovine fibrinogen (Fluka, in 0.1 M phosphate buffer, pH 7.2), then rinsed 10 times with water and dried under a stream of N_2.

2.7. Ellipsometry

A Rudolph Auto EL-II Null-Ellipsometer (New Jersey, USA) equipped with a He–Ne laser ($\lambda = 632.8$ nm) with an angle of incidence fixed at 70.0° was used to determine the thicknesses [23] of the gold films and those of the adsorbed cellulose and proteins. The incident laser beam covered an area of approximately 3 mm^2 and the samples were measured at a number of different points. For the interpretation of the ellipsometric data a multilayer model [28] was used, assuming refractive indices n for Si of $3.858 - i0.018$ [24], for SiO_2 of 1.462, for Au of $0.14 + i3.14$ [25], for TSHP of 1.50 and for BSA and fibrinogen of 1.518 [26,27]. If the adsorbed film is regarded as a homogeneous layer, the thickness can be obtained by standard procedures [22,23,28].

2.8. Contact angles

Contact angles of water drops (4 μl) were measured at room temperature immediately after the surface modification by a standard technique [29]. Measurements were taken on both sides of water drops 30 s after they were applied to the surface. Each reported value is the average of three measurements taken at different spots on the film.

2.9. FTIR-External reflectance spectroscopy (ERS)

FTIR-External Reflectance Spectroscopy (ERS) of the adsorbates was performed at room temperature with a Bruker IFS 28 spectrometer by reflection of the incident beam at an incident angle of 84° using p-polarized radiation. The detector was mercury cadmium tellury (MCT) type, cooled

with liquid N_2. Spectra were recorded at 2 cm^{-1} resolution and are the average of 500 individual scans. Reference spectra were obtained from fresh gold mirrors.

2.10. Atomic force microscopy (AFM)

AFM investigations were carried out with a universal instrument from Park Scientific (Sunnyvale, CA) equipped with a homebuilt head containing laser deflection detection system. Measurements were made in the contact mode in air at room temperature. V-Shaped silicon nitride cantilevers with sharpened pyramidal tips and force constants between 0.03 and 0.1 N/m were applied. The total force load was approximately 5 nN (including capillary forces). All AFM images represent unfiltered original data and are displayed in a linear gray scale.

2.11. XPS (X-ray photoelectron spectroscopy)

XPS experiments were performed using an ESCALAB-5 electron spectrometer (VG Scientific, East Grinstead, UK) in a UHV-system with a base pressure of ca. 10^{-1} mbar. The photoelectrons were excited in a sample area of about 50 mm^2 by non-monochromatized Mg Kα-radiation at a power of 100 W. The kinetic energies were measured by a 150° hemispherical energy analyzer operated in the constant analyzer energy mode (CAE) using a pass energy of 20 eV for elemental spectra and a resolution of 1.3 eV for the Au $4f_{7/2}$ photopeak. In relation to the normal samples the photoelectron take-off angles for the gold and silicon substrates were 40° and 0°, respectively. The binding energy scale was calibrated using a value of 285.0 eV for the contamination C 1s photopeak and controlled by means of the well known photopeaks of metallic Cu, Ag, and Au. For the analysis of multiple peaks in XPS spectra the VGX 900 software permits simultaneous fitting of up to six gaussian components with adjustable Lorentzian line shape contributions and asymmetries.

3. Results and discussion

3.1. Synthesis

The synthesis of the cellulose derivative TSHP is shown schematically in Fig. 1. First cellulose, activated by treatment with NaOH, was reacted with allyl glycidyl ether. Allyl-oxy-2-hydroxypropyl substituents were introduced mainly at position O-6 of the cellulose. In the water soluble product AHP, a degree of substitution (DS) of 0.7 was achieved. Preferential substitution by the allyl glycidyl ether at O-6 was attributed to the greater accessibility of the primary hydroxyl groups in relation to the secondary. Similar regioselectivities were found with other bulky reagents, e.g. trityl chloride [30,31]. In aqueous solution at high temperatures (80–100°C) there was addition of tetrathionate to some of the double bonds of AHP. The resulting polymer TSHP contained 10% 6-*O*-(2,3-bis(thiosulfato)propyl-oxy-2-hydroxy-propyl)-anhydroglucose units, about 30% unmodified anhydroglucose units, and 60% unreacted 6-*O*-(allyl-oxy-2-hydroxy-propyl)-anhydroglucose units, distributed statistically along the chain. The extent of conversion in the addition reaction was determined from the decrease in the ^{1}H-NMR signal intensities of the allyl group relative to those of the anhydroglucose unit, and also by elemental analysis. The product TSHP displayed IR bands at 601 and 408 cm^{-1} (Fig. 2a) characteristic of C–S–S moieties. The product was readily soluble in water and showed no tendency to precipitate.

Alkyl thiosulfates had previously been synthesized by nucleophilic attack of thiosulfate ions on alkyl halides or tosylates, or by the addition of thiosulfate to activated double bonds [32,33]. The alternative described here of adding tetrathionate across double bonds appears to be an interesting new approach to the generation of vicinal bisthiosulfates. The addition may begin with homolytic cleavage of the S–S bond of the tetrathionate (Eq. (1)).

Each of the resulting thiosulfate radicals should then add to the C=C double bond (Eq. (2)). This hypothesis is supported by our observation that reaction occurs only at

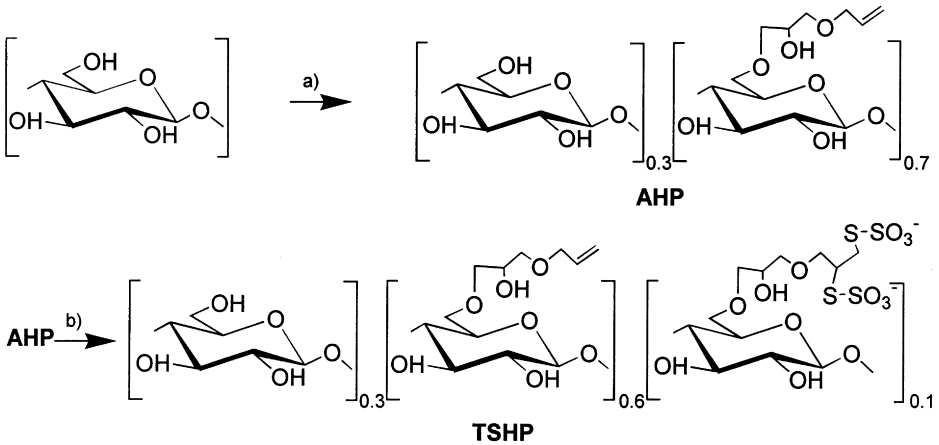


Fig. 1. Synthesis of TSHP. (a) Allyl glycidyl ether/NaOH; (b) $K_2S_4O_6/H_2O$.

high temperatures (90–100°C). The low rate of addition is evidence against a radical chain mechanism.

$$^{-}O_3S-S-S-SO_3^{-} \rightarrow 2\,^{-}O_3S-S\cdot \quad (1)$$

$$2O_3S-S\cdot + R-CH{=}CH_2 \rightarrow R-CH(S-SO_3^{-}) \quad (2)$$

$$-CH_2S-SO_3^{-}$$

The formation of thiosulfate salts has the advantage that the educt, e.g. the allyl derivative AHP, is very unreactive. In contrast to alkyl halide or tosylate groups the remaining allyl groups should not interfere with biological components such as proteins. A second advantage is the low tendency of TSHP to crosslink by forming disulfide bridges. Stable aqueous solutions of TSHP were obtained.

3.2. Immobilization of TSHP on Au

3.2.1. Film thickness by null-ellipsometry

The thickness of the TSHP adsorbed onto gold substrates from dilute aqueous solution at room temperature was investigated by null-ellipsometry. The TSHP layer had a mean thickness of 4 ± 1 nm, which was independent of concentration within the studied range. The thickness measured for the TSHP films on gold is similar to the values reported for other sulfur-derived polymers with comparable molecular weights [15,16].

The thickness indicates strong TSHP adsorption, and is probably due to chemical bonding between the functional groups and the gold surface (this will be discussed in detail with the XPS results). For comparison, the adsorption of TSHP onto bare silicon wafers was investigated under the same conditions. This gave only a very thin layer (0.4 nm), showing the low affinity of TSHP for SiO_2.

To obtain information about the adsorption kinetics, bare gold substrates were immersed in aqueous solutions of TSHP for 5, 15, 30, 60 and 240 min. After the first hour there was no further change in thickness. A more accurate kinetic study would require an ellipsometry set up for in situ measurements.

The adsorption of TSHP from aqueous solution onto gold was found to be irreversible. Gold substrates coated with TSHP were immersed in pure water for 1 week, then dried and investigated again by null-ellipsometry. There had been no significant reduction in layer thickness.

3.2.2. FTIR-ERS

This can yield information about orientation and chain conformation in adsorbed monolayers, as is well reported in the literature [5–9]. A typical polarized infrared reflectance spectrum of TSHP cellulose adsorbed onto gold (Fig. 2b) was compared with its bulk transmission spectrum (KBr pellet, Fig. 2a). The TSHP film has finer band shapes than TSHP in bulk, but the characteristic bands of cellulose are present in both spectra: in the 3500–3200 cm^{-1} region (OH vibrational stretching); at 2930 cm^{-1} and 2850 cm^{-1} (symmetrical and asymmetrical CH stretching); and in the 1200–850 cm^{-1} region (C–O and C–C stretching vibrations of the glucopyranose ring). In the latter complex region the shoulder at 1156 cm^{-1} was assigned to the asymmetric stretching vibration of the C–O–C moiety bridging two adjacent glucose units [34].

3.2.3. Atomic force microscopy (AFM)

AFM analysis was performed before and after the adsorption of TSHP, in order to study how the TSHP chains assembled on the gold surface. Comparable images were obtained for different regions within the same sample and for different samples. Typical topographic AFM images of bare gold and of TSHP cellulose coated gold are shown in Fig. 3a,b. For the bare gold layers characteristic islands [35] with a mean diameter of 32 ± 3 nm and a mean roughness (rms) of 4.9 ± 0.2 nm were found in areas of 0.1 μm^2. After adsorption of TSHP the surfaces exhibited similar islands, with practically the same average diameter (34 ± 3 nm) and roughness (4.7 ± 0.2 nm). Thus deposition of the TSHP monolayer produced no observable change in the AFM images. The cellulose chains might be too soft to be visualized in this AFM mode.

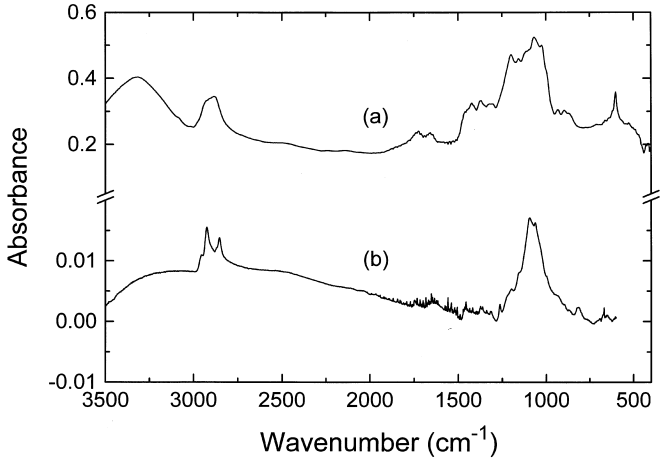


Fig. 2. (a) FTIR spectra of bulk TSHP in KBr pellets; (b) Polarized FTIR-ERS spectra of TSHP film adsorbed onto gold.

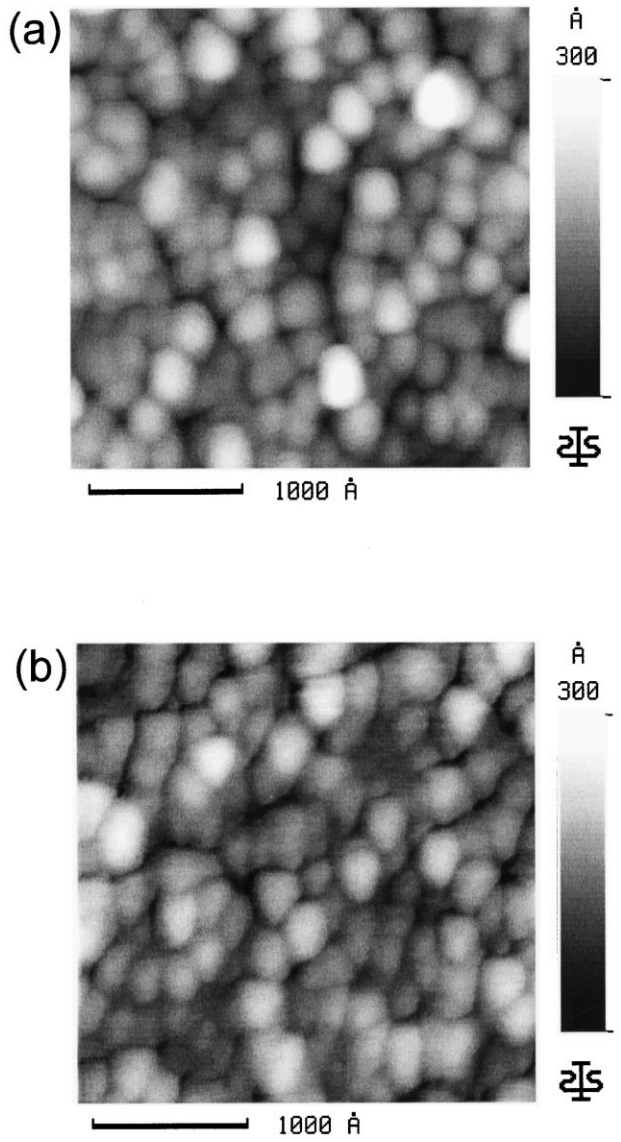


Fig. 3. (a) AFM topographic image of bare gold evaporated onto silicon wafer; (b) AFM topographic image of TSHP adsorbed onto gold.

3.2.4. Contact angle measurements

The bare gold substrates used were quite hydrophobic. The advancing and receding contact angles of water on the gold substrates were $\theta_a = 89 \pm 2°$ and $\theta_r = 55 \pm 2°$, respectively. Adsorption of TSHP reduced the contact angles to $\theta_a = 53 \pm 2°$ and $\theta_r = 16 \pm 2°$. This increase in hydrophilicity was probably due to the many hydroxyl groups in the cellulose chains. The similarity of the hystereses of the contact angles, $\theta_a - \theta_r = 34°$ for Au and $\theta_a - \theta_r = 37°$ for TSHP on Au, showed that surface roughness was nearly identical for the two surfaces, as already observed by AFM analysis.

3.2.5. X-Ray photoelectron spectroscopy

XPS spectra of the S 2p photopeak obtained for TSHP on silicon wafer showed two species: sulfide at 163.5 ± 0.4 eV and sulfate at 168.2 ± 0.4 eV (Fig. 4a). These results were interpreted as a weak physisorption of the TSHP. Corresponding examination of the S 2p region for TSHP adsorbed on gold revealed some interesting features, as shown in Fig. 4b. In this case only the photopeak at 162.3 ± 0.4 eV could be detected. The binding energy is very similar to that found for metallic sulfides [36]. In both spectra the S 2p doublets are unresolved, apparently because of broadening by surface charging and the very low intensity of the photopeaks.

The chemisorption of organic sulfides [2–5] and disulfides [6,8] onto gold occurs by the formation of gold thiolates. The Au–S bond is known to be very strong, with a typical bond energy of about 40–45 kcal/mol [2,37]. In the case of the chemisorption of disulfides, homolytic cleavage of the sulfur–sulfur bond proceeds by direct oxidative addition to the gold [6,7]. The chemisorption of TSHP onto gold may be explained by an analogous mechanism. We suggest that the S–S bonds in the $S-SO_3^-$ groups are homolytically cleaved. The free thiol radicals in the cellulose chain then oxidise the gold leading to covalent S–Au bonds. Sulfite free radicals are liberated into solution and dimerize to form dithionate, as illustrated schematically in Eqs. (3)–(5) and Fig. 5. The amount of dithionate released ($\sim 10^{-10}$ mol/cm^2) is too small for detection by conventional analytical methods.

$$R - S - SO_3^- \rightarrow R - S\cdot + \cdot SO_3^- \quad (3)$$

$$R - S\cdot + Au^0 \rightarrow R - S^- Au^+ \quad (4)$$

$$2\cdot SO_3^- \rightarrow S_2O_6^{2-} \quad (5)$$

On the basis of an interatomic gold lattice spacing [7,38] of 2.9 Å and standard lengths and angles for the C–C and C–S bonds, a six-membered Au sulfide ring, as shown in Fig. 5, would be sterically feasible.

In this model most of the thiosulfate groups which are attached statistically along the TSHP chain bond to the gold surface, while the non-functionalized chain segments in

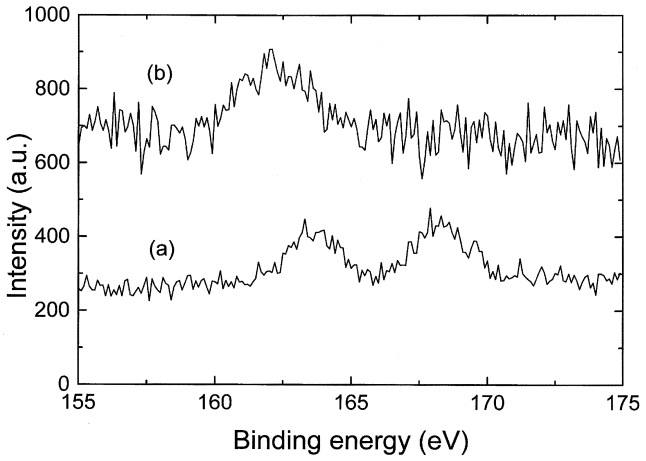


Fig. 4. (a) S 2p XPS spectra of TSHP cast on silicon wafer (b) S 2p XPS spectra of TSHP adsorbed onto gold.

between dangle as loops into the solution. If all thiosulfate groups reacted, the loops would have an average length of 10 anhydroglucose units, corresponding at a unit length of about 0.6 nm to a contour length of 6 nm, or a loop height of 3 nm. This estimate agrees closely with the measured value. Since there is no strong evidence for orientation of the cellulose, the TSHP layers are likely to be rather soft. Their structure is therefore quite different from the quasi-crystalline [2,5,7] structure of alkanethiols adsorbed on gold. The use of a polyfunctional polymer like TSHP rather than a monofunctional alkanethiol is advantageous because of the high cooperativity of the immobilization. The absence of crystallinity can also be helpful in providing a high microscopic homogeneity. Perspectives on amorphous so-called 'fuzzy nanoassemblies' of polymers have recently been outlined [39].

3.3. Protein adsorption on TSHP monolayers

The adsorption of blood proteins (BSA, fibrinogen) was investigated by null-ellipsometry, AFM and FTIR-ERS. We chose BSA for our adsorption studies because it is one of the most abundant proteins in blood, and fibrinogen because of its major contribution to the blood clotting process.

3.3.1. Adsorption of BSA onto TSHP layers

The thicknesses of the protein films adsorbed onto TSHP layers were determined by null-ellipsometry. A typical mean thickness of 0.4 ± 0.1 nm was found for BSA. Since it is a globular protein with a high average molecular weight (about 70 000 g/mol), and given that ellipsometry yields only a mean value, such a low thickness reading must imply a highly heterogenous film. This supposition was confirmed by studies with AFM. In Fig. 6, a typical image shows aggregates with average width 600 nm and average height 250 nm sparsely distributed across the surface. These aggregates covered only 6% of the total surface area. Their average volume was determined from a combination of many AFM images by approximating their shape to spherical sections. Corrections were made for the effects of tip broadening. The volume figure obtained was divided by the surface area, giving a rough estimate of the mean thickness of the adsorbed BSA of 0.9 nm, or the same order of magnitude indicated by null-ellipsometry.

3.3.2. Adsorption of fibrinogen onto TSHP layers

Fibrinogen was adsorbed onto TSHP-coated gold surfaces more strongly than BSA. Ellipsometric measurements gave a mean thickness of 1.4 ± 0.2 nm. Fibrinogen [40] (average molecular weight 400 000 g/mol) is a fibrilar protein of 65 nm length and 2 nm diameter. Fig. 7 shows a typical AFM image of the adsorbed fibrinogen. Large 'worm-like' aggregates can clearly be seen. On average they are 500 nm wide, 150 nm high and 2000 nm long, and they cover 10% of the total surface area. They also occur on the surface more frequently than the BSA aggregates. A mean thickness of 2.6 nm for the adsorbed fibrinogen was estimated from AFM images, using a similar procedure to that described above for BSA. Again, in view of experimental error and the geometrical assumptions which were made, there is reasonable agreement between the thicknesses estimated from AFM analysis and null-ellipsometry.

The adsorption of fibrinogen onto TSHP layers was also investigated by FTIR-ERS. The resulting spectra were compared with those obtained for pure TSHP films (Fig. 8). The characteristic cellulose absorption bands are present in both spectra. However, the fibrinogen coated sample shows some additional features (Fig. 8b). A strong band at 1654 cm^{-1} and a weak band at 1545 cm^{-1} were assigned to the CO stretching vibration of the peptide carbonyl group (amide I) and the NH bending vibration of the peptide (amide II), respectively. The increased relative intensity of the broad

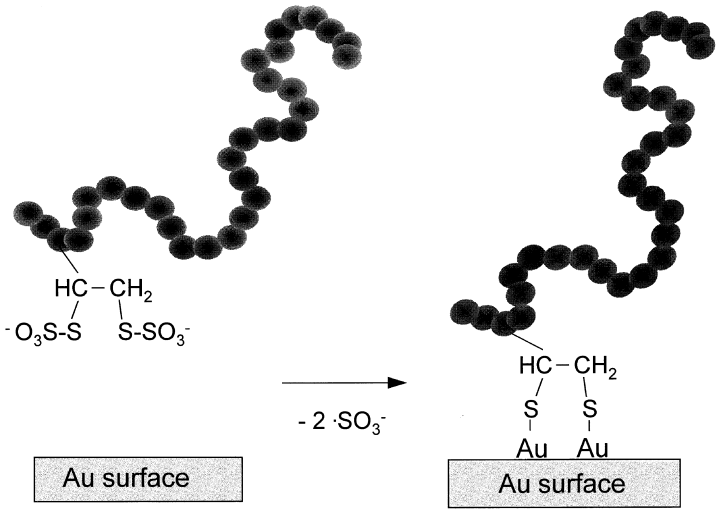
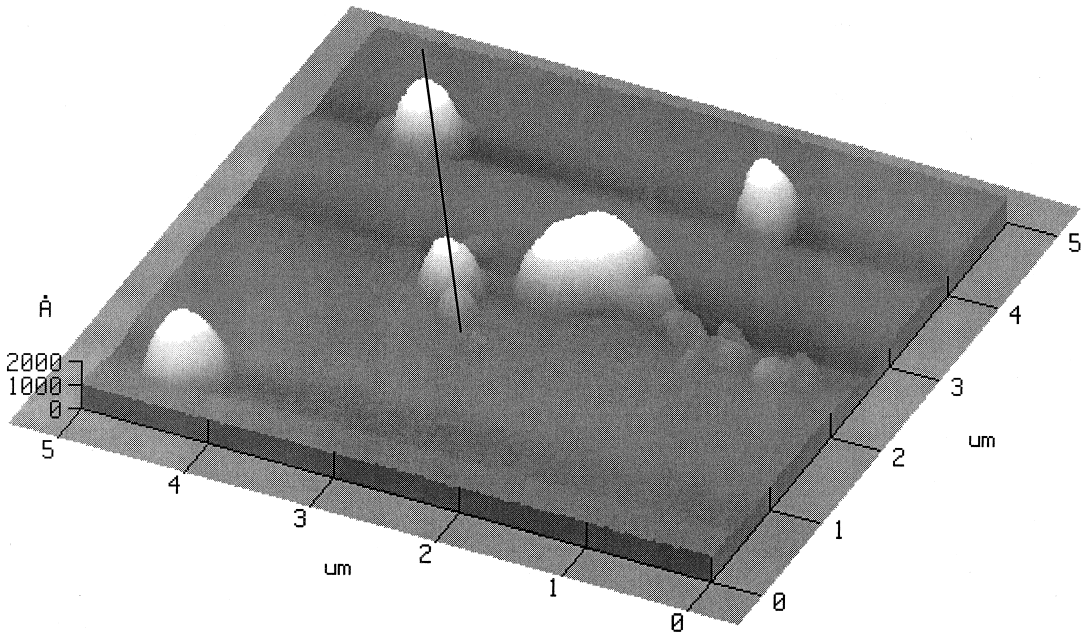


Fig. 5. Schematic representation of the adsorbed TSHP chains onto gold surface.

adsorption between 3500 cm^{-1} and 3200 cm^{-1} is attributed to the NH stretching vibration.

The principal forces driving the adsorption of proteins onto solid surfaces are hydrophobic and ionic interactions, combined with entropy gains due to conformational changes in the proteins [41]. The affinity of BSA and fibrinogen for TSHP coated gold seems to be very low. The average protein layer thicknesses (BSA $d = 0.4$ nm, fibrinogen $d = 1.4$ nm) were small and there was effectively no adsorption over 90–95% of the surface. In comparison the adsorption of blood proteins onto uncoated gold (BSA $d = 2$ nm, fibrinogen $d = 8$ nm) or hydrophobic surfaces (BSA $d = 4$ nm, fibrinogen $d = 28$ nm) [26,27] is quite facile. The relatively poor adsorption of proteins onto TSHP coated surfaces is probably due to the hydrophilicity of cellulose. The affinity of fibrinogen for oligo(ethylene glycol)-terminated SAMs adsorbed on silver and gold was recently reported [42]. The advancing water contact angles measured on these monolayers were similar to those found on TSHP. The SAMs showed either no interaction with fibrinogen (on gold) or only a weak interaction (on silver).

The AFM images in Figs 6 and 7 show that the blood proteins form isolated aggregates on the TSHP film. The influence of concentration, time of immersion and surface roughness on the nucleation of these aggregates will be the subject of further investigations.



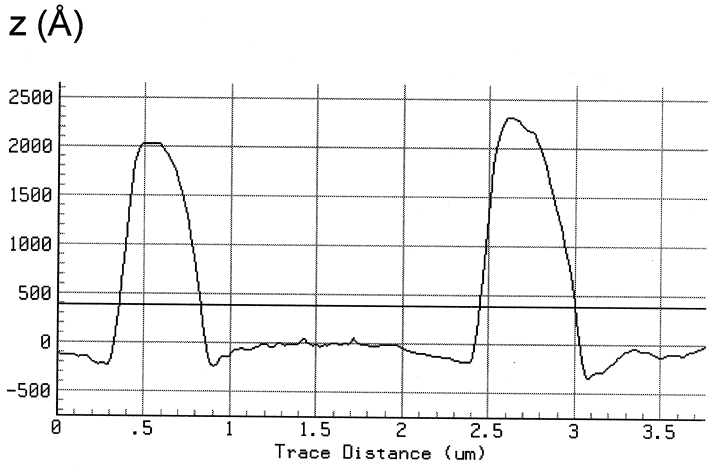


Fig. 6. AFM image of BSA adsorbed onto TSHP coated gold surface with respective section analysis.

4. Conclusions

TSHP is a new water-soluble cellulose derivative. On average 10% of repeat units in the chain bear thio moieties able to anchor the polymer to the gold surface. From aqueous solution TSHP appears to form dense ultrathin cellulose layers on gold surfaces. A chemisorption model has been proposed based on the homolytic cleavage of the thiosulfato groups leading to cellulose-thiol free radicals which then oxidize the gold. XPS measurements detected only sulfide on the metal surface. There was no evidence of residual thiosulfate groups in the monolayer. The multi-site attachment of the cellulose chain by formation of gold thiolates guarantees a high film stability. Other metals which form covalent sulfides, such as silver and copper, might also accept coating by TSHP. This could improve their biocompatibility or prevent corrosion.

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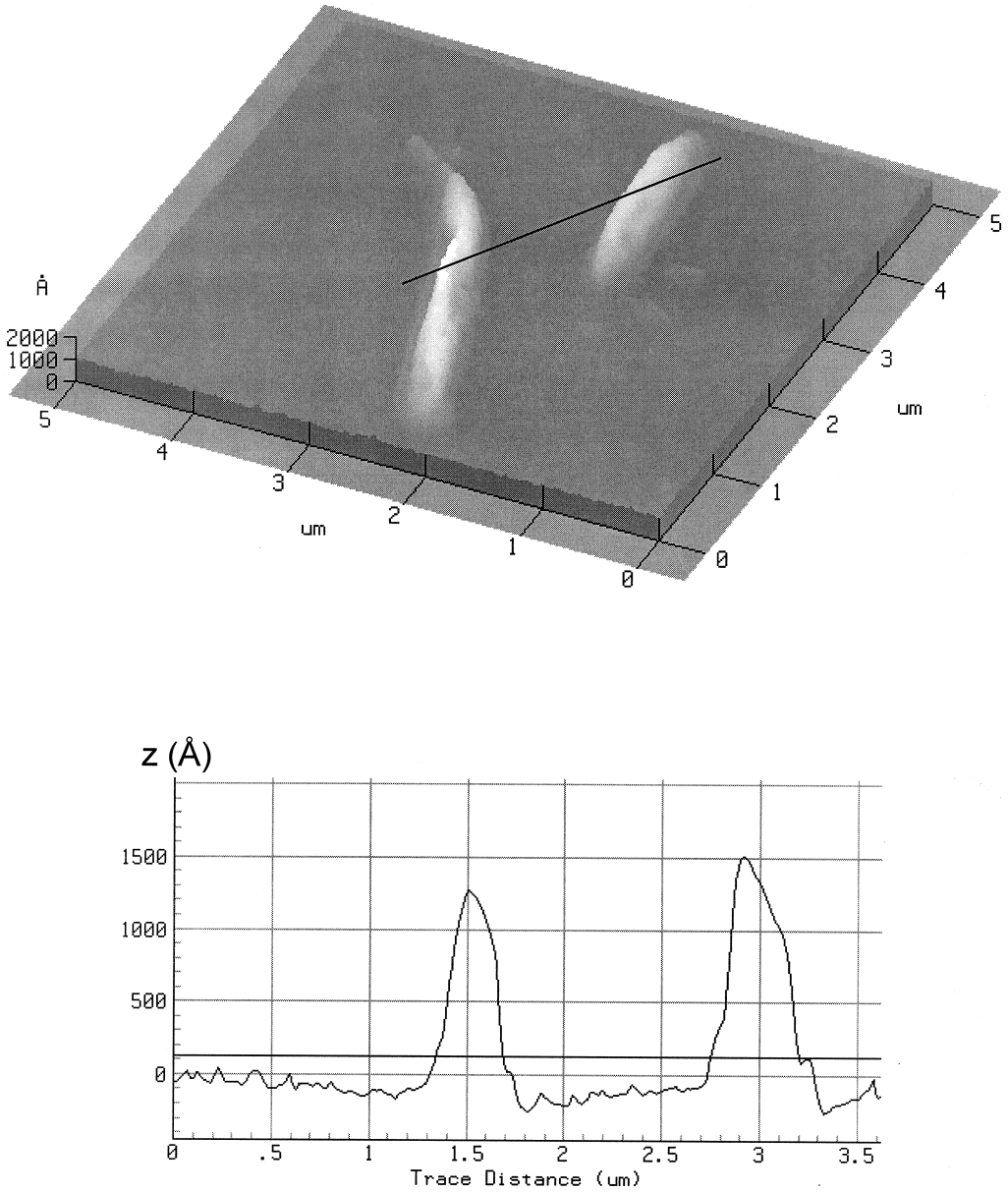

Fig. 7. AFM image of fibrinogen adsorbed onto TSHP coated gold surface with respective section analysis.

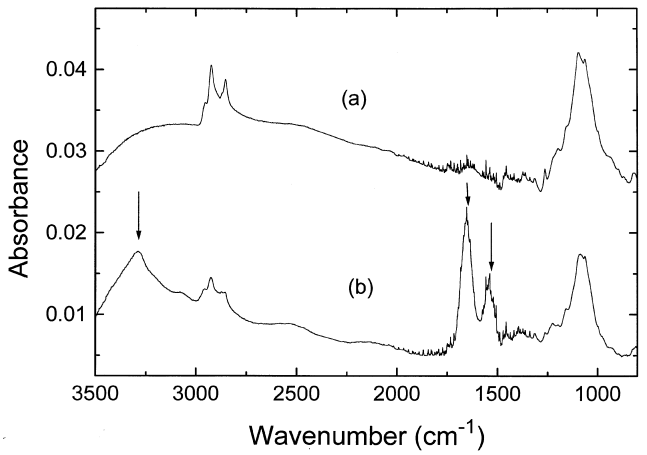

Fig. 8. (a) Polarized FTIR-ERS spectra of TSHP film adsorbed onto gold, (b) Polarized FTIR-ERS spectra of fibrinogen adsorbed onto TSHP film. The arrows indicate the typical bands for the peptide.

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